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The Self-reported Use of Probiotics is Associated with Better Glycaemic Control and Lower Odds of Metabolic Syndrome and its Components in Type 1 Diabetes

Aila J Ahola[1,2,3], Valma Harjutsalo[1,2,3,4], Carol Forsblom[1,2,3], Riitta Freese[5], Sari Mäkimattila[6] and Per-Henrik Groop[1,2,3,7*]

[1]*Folkhälsan Institute of Genetics, Folkhälsan Research Center, Helsinki, Finland*

[2]*Abdominal Center Nephrology, University of Helsinki and Helsinki University Central Hospital, Helsinki, Finland*

[3]*Research Programs Unit, Diabetes and Obesity, University of Helsinki, Finland*

[4]*National Institute for Health and Welfare, Diabetes Prevention Unit, Helsinki, Finland*

[5]*Division of Nutrition, Department of Food and Environmental Sciences, University of Helsinki, Finland*

[6]*Abdominal Center Endocrinology, University of Helsinki and Helsinki University Central Hospital, Helsinki, Finland*

[7]*Department of Diabetes, Central Clinical School, Monash University, Melbourne, Victoria, Australia*

[*]**Corresponding author:** Per-Henrik Groop, Folkhälsan Research Center, University of Helsinki, Biomedicum Helsinki C318b, PO Box 63, 00014, Finland, Tel +358 2941 25459; E-mail: per-henrik.groop@helsinki.fi

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Abstract

In type 1 diabetes, various metabolic disturbances are frequently observed. Importantly, these may negatively affect individuals' long-term health outcomes. The use of probiotics has, in other populations, been beneficially associated with many of these risk factors. We, therefore, assessed the cross-sectional associations between the use of probiotics-containing food products or supplements and various health markers in a large population of individuals with type 1 diabetes.

Included were 1039 individuals (mean age 46 ± 14 years, 45% men) with type 1 diabetes and without end-stage renal disease. Based on the entries in the diet questionnaire and the food record, participants were divided into those using (Probiotics^{yes}) and not using (Probiotics^{no}) probiotics-containing products. Various standard health markers, such as weight, waist circumference, blood pressure, blood lipids, and HbA_{1c}, were measured during the study visit.

In all, 403 (39%) individuals reported using probiotics. Adjusted with potential confounders, the rate of overweight/obesity, body mass index, and waist-to-hip ratio were higher in the Probiotics^{no} group. Moreover, the odds of metabolic syndrome, and its waist, blood pressure, HDL-cholesterol, and triglyceride components were higher amongst those not using probiotics. In the normal-weight individuals, using probiotics was associated with significantly better glycaemic control.

Using probiotics-containing food products or supplements may beneficially affect many of the traditional risk factors related to the diabetic complications. Randomized controlled trials are required to verify these observations.

Keywords: Glycaemic control; Lipid profile; Metabolic syndrome; Obesity; Probiotics; Type 1 diabetes

Introduction

Diabetes is characterized by pathologically increased plasma glucose levels [1]. Importantly, hyperglycaemia is an independent risk factor for the development of vascular complications [2] and mortality [3]. In addition to glycaemia, however, various other metabolic abnormalities are frequently present in diabetes. In one Finnish study, 55% of individuals with type 1 diabetes, for example, failed to meet the LDL-cholesterol concentration goals [4]. We have also previously shown, in the Finnish Diabetic Nephropathy (FinnDiane) Study that, of the individuals with type 1 diabetes and without diabetic nephropathy, only 35% reached the target levels for the LDL-cholesterol concentration [5]. In the same study, of the diabetic nephropathy-free individuals with type 1 diabetes, 45% were observed to reach the blood pressure targets, while the respective figure for those with diabetic nephropathy was only 19% [5]. In one review, the reported rates of obesity or overweight, among adult individuals with type 1 diabetes, ranged between 44% and 78% [6]. These risk factors, especially the clustering of multiple risk factors, are associated with increased risk of various vascular complications [7], detrimental to the longevity of the affected individuals [8].

The importance of managing these risk factors is stressed in various national and international treatment guidelines [9-11]. According to these guidelines, lifestyle management is a fundamental aspect in diabetes care, and includes components such as nutrition therapy, physical activity, and smoking cessation. Beyond lifestyle modification, however, pharmacological approaches are frequently required in order to achieve the treatment goals.

The role of the intestinal microbiome, in modifying various metabolic risk factors, has in recent years gained increasing interest. In the human intestine, Bacteroidetes and Firmicutes form the majority of the total microbial population [12]. The relative abundance of these phyla has, however, been observed to differ between lean and obese, with higher proportion of Firmicutes to Bacteroidetes in the obese [13]. Moreover, lower bacterial richness in the human gut has been associated with markedly higher body weight and fat mass [14]. In addition to the body weight-related variables, the quality of the intestinal ecosystem has been associated with systemic inflammation, plasma lipids, glycaemia, and blood pressure [14-16].

Probiotics-containing food products or supplements have been used to favourably modify the gut's microbial composition [17]. According to the definition, published by the joint Food and Agriculture Organization/World Health Organization Expert Consultation, probiotics are "live microorganisms which when administered in adequate amounts confer a health benefit on the host" [18]. These health benefits have been investigated in various populations. For example, in a meta-analysis of nine randomized controlled trials among adults with or without hypertension, probiotic interventions were observed to reduce systolic and diastolic blood pressure [19]. In another meta-analysis of individuals with high, borderline high, and normal cholesterol levels, reduction in total and LDL-cholesterol concentrations, following probiotics-intervention, was reported [20]. Finally, significant lowering of HbA_{1c}, fasting blood glucose, and fasting plasma insulin concentrations, and amelioration of insulin resistance have been seen after interventions with probiotics-containing products in type 2 diabetes [21,22]. To the best of our knowledge, the health effects of probiotics amongst individuals with type 1 diabetes, a population at high risk of vascular complications, have not been investigated.

The aim of the current study was to assess the cross-sectional associations between reported use of probiotics-containing food products or supplements and selected health parameters in a large population of well-defined individuals with type 1 diabetes.

Materials and Methods

Study subjects

Study subjects were individuals with type 1 diabetes participating in the FinnDiane Study. Type 1 diabetes was presumed if age at diabetes onset was below 35 years and if permanent insulin treatment was initiated within one year of the diagnosis. Included, in the current analyses, were all individuals who had completed the diet questionnaire within two years from the study visit [median (interquartile range) interval between the study visit and the completion of diet questionnaire was 6 (0-61) days]. Those with end-stage renal disease (undergoing dialysis or with a kidney transplantation) were excluded from the analyses. In all, data from a total of 1039 individuals (mean age 46 ± 14 years, 45% men) were included. The study protocol was approved by the Ethics Committee of the Helsinki University Central Hospital. Study subjects provided written informed consent prior to participation.

Clinical measurements

During the FinnDiane study visit, participants' height and weight were measured in light clothing, and body mass index was calculated (BMI; kg/m^2). Overweight and obesity were defined as BMI ≥ 25 kg/m^2 and ≥ 30 kg/m^2, respectively. Waist and hip circumferences were measured using an inelastic tape measure. Following a minimum of ten-minute rest, blood pressure was measured while seated. A second measure was performed with a two-minute interval. Mean of the two measurements was used in the analyses. HbA_{1c} was determined locally using standardized assays. Serum lipid and lipoprotein concentrations were measured centrally at the research laboratory of the Helsinki University Central Hospital. Serum triglyceride concentration was measured using a Konelab 60i analyser (Thermo Fisher Scientific Inc., Waltham, MA, USA), and serum HDL-cholesterol concentration was measured with a HTS 7000 plus Bio Assay Reader (Perkin Elmer Inc., Waltham, MA, USA). Renal status was assessed based on the urinary albumin excretion rate (AER) in at least two out of three timed 24-h or overnight urine collections. Diabetic nephropathy was assumed when AER ≥ 200 μg/min or ≥ 300 mg/24 h. Serum hs-CRP concentration was measured by immunoassay (Modular analyzer, Roche). Individuals with hs-CRP values above 10 mg/l were excluded from the hs-CRP analyses, as they likely had an acute infection. Smoking was self-reported. The attending physician recorded medication use on a standardized questionnaire. From these questionnaires, data regarding antihypertensive and blood lipid medication were collected. Information on the use of antibiotics within 6 months prior to the study visit was obtained from the Drug Prescription Register of the Social Insurance Institute of Finland.

Metabolic syndrome

The criteria established by International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity was used to define the metabolic syndrome [23]. Accordingly, metabolic syndrome exists when the minimum of three of the following components coexist: waist circumference ≥ 94 cm in men and ≥ 80 cm in women, triglyceride concentration ≥ 1.70 mmol/l (or medication to lower triglyceride concentration), HDL-cholesterol concentration <1.00 mmol/l in men and <1.30 mmol/l in women (or medication to increase HDL-cholesterol concentration), blood pressure ≥ 130/85 mmHg (or antihypertensive medication), and fasting blood glucose concentration ≥ 6.11 mmol/l. Diagnosed with type 1 diabetes, all patients were defined to fulfil the hyperglycaemia criterion.

Dietary intake

Data on the participants' dietary intake were collected using two separate methods, as previously described [24]. In short, at the study visit, participants were provided with a validated [25] diet questionnaire. With this questionnaire, we aimed at obtaining an overview of the subjects' dietary intake. Amongst others, we enquired the types of milk, bread, spread, and cooking fat typically consumed. As part of the questionnaire, a short food frequency questionnaire was included. Of interest for the current study, we also queried about the use of probiotics-containing food products or supplements over the past month (no/yes, if yes provide the trade name and amount or dose consumed). Upon returning the diet questionnaire, subjects were sent a 3-day exercise and food record. In this record, along with reporting physical activity and insulin administration, all foods and drinks consumed over the allocated consecutive days (two weekdays and one weekend day), were recorded. Another 3-day food record was completed with a 10-week interval. Up to two reminders were sent to

non-responders. Dietary composition of the food record entries was analysed using the Diet 3.2 software (version 1.4.6.2, AIVO, Turku, Finland) and, from August 2014 onwards, AivoDiet software (version 2.0.2.3, AIVO, Turku, Finland), both based on the Finnish National Food Composition Database. In the analyses the health markers of individuals, reporting (Probiotics^{yes}) and not reporting (Probiotics^{no}) the use of probiotics-containing food products or supplements, either in the diet questionnaire or in the food record, were compared.

Physical activity

Leisure-time physical activity (LTPA) was estimated using a validated questionnaire, as previously described [26]. In this form, participants reported frequency, single session duration, and intensity of a number of most typical physical activities. Based on these reports, weekly LTPA was calculated as the metabolic equivalent of task hours (MET h/week).

Statistical analyses

Descriptive statistics are reported as percentages for categorical data, mean ± standard deviation for continuous normally distributed data, and median (interquartile range) for continuous non-normally distributed data. The respective between-group comparisons were done using Chi-squared test, independent samples t-test, and Mann-Whitney U-test. The difference in the outcome variables, between the two probiotics-groups, were further tested in the multivariable models. For dichotomous outcome variables, logistic regression analyses were applied, while with continuous variables, generalised linear regression analyses were conducted. Analyses of waist circumference and HDL-cholesterol concentration were conducted separately for men and women, as there are sex-specific cut off values for these variables also in the definition of the metabolic syndrome. HbA_{1c} analyses were conducted separately for normal-weight and overweight or obese individuals, as there is indication that probiotics may have distinctive effects depending on the adiposity [27]. Confounders were selected to the multivariable models if they significantly differed between the two groups or probiotics-consumption, or if they were considered to significantly contribute to the outcome variable (e.g., lipid lowering medication in blood lipid analyses, and antihypertensive medication in blood pressure analyses). A two-tailed P value <0.05 was considered statistically significant. All data were analysed using IBM SPSS Statistics for Windows, Version 22.0 (IBM Corp, Armonk, NY, USA).

Results

Of the 1039 participants, 403 (39%) reported using probiotics-containing food stuffs or supplements (Probiotics^{yes}, Table 1). In this group, the proportion of men and those with diabetic nephropathy was lower. No differences were observed, between the groups, in mean age, the frequency of antibiotic use within 6 months prior to the study visit, the frequency of smoking, or the level of physical activity.

Table 1: Participant characteristics divided by the reported use of probiotics.

	Probiotics^{no} n=636 (61%)	Probiotics^{yes} n=403 (39%)	P
Men, %	51	35	<0.01
Age, years	46 ± 14	46 ± 13	0.45
Use of antibiotics[a], %	30	32	0.439
Diabetic nephropathy, %	14	9	0.008
Smoking, %	14	12	0.565
Physical activity, MET h/week	12 (2-27)	13 (1-28)	0.641
Dichotomous outcome variables			
Overweight or obese, %	58	49	0.006
Metabolic syndrome, %	70	58	<0.001
MS-Blood pressure component, %	81	72	0.002
MS-HDL-cholesterol component, %	47	35	<0.001
MS-TG component, %	47	33	<0.001
MS-waist component, %	58	51	0.032
Continuous outcome variables			
HbA_{1c} (NW), mmol/mol	64 (57-75)	62 (54-69)	0.01
HbA_{1c} (OW/OB), mmol/mol	65 (57-74)	66 (60-74)	0.277
HbA_{1c} (NW), %	8.0 (7.4-9.0)	7.8 (7.1-8.5)	0.01
HbA_{1c} (OW/OB), %	8.1 (7.4-8.9)	8.2 (7.6-8.9)	0.277

BMI, kg/m^2	25.9 (23.3-28.6)	25.0 (23.0-27.8)	0.011
Waist-to-hip ratio	0.89 (0.83-0.96)	0.87 (0.82-0.92)	<0.001
Waist circumference (men), cm	94 (86-102)	92 (84-98)	0.041
Waist circumference (women), cm	84 (77-92)	82 (75-91)	0.093
Systolic blood pressure, mmHg	136 (126-149)	133 (124-147)	0.032
Diastolic blood pressure, mmHg	77 (71-84)	76 (70-83)	0.09
Total cholesterol, mmol/l	4.6 (4.0-5.2)	4.6 (4.1-5.1)	0.582
HDL-cholesterol (men), mmol/l	1.42 (1.17-1.67)	1.50 (1.21-1.79)	0.072
HDL-cholesterol (women), mmol/l	1.64 (1.35-1.96)	1.72 (1.45-2.09)	0.026
TG, mmol/l	0.99 (0.76-1.42)	0.91 (0.69-1.23)	<0.001
TG-HDL-cholesterol ratio	0.65 (0.43-1.08)	0.56 (0.37-0.82)	<0.001
hs-CRP, mg/l	1.11 (0.54-2.53)	0.97 (0.51-2.27)	0.212

Data are presented as frequencies (%) for categorical variables, mean± standard deviation, or median (inter-quartile range). Between-group comparisons are conducted using Chi squared test, independent samples t-test, and Mann-Whitney U-test, respectively. [a]Use of antibiotics within 6 months prior to the study visit. MET, metabolic equivalent of task, a measure of physical activity; MS, metabolic syndrome; HDL, high density lipoprotein; TG, triglyceride; NW, normal-weight (BMI <25 kg/m^2); OW/OB, overweight or obese (BMI ≥ 25 kg/m^2); BMI, body mass index; hs-CRP, high-sensitivity C-reactive protein.

Compared to those reporting the use of probiotics, various overweight-related measures were more prevalent amongst those not using probiotics (Table 1). For example, BMI and waist-to-hip ratio were higher in the Probiotics^{no} group. Moreover, the prevalence of overweight or obesity, and the frequency of fulfilling the waist component of the metabolic syndrome were higher in the Probiotics^{no} group. In addition, the metabolic syndrome and all the remaining components of the metabolic syndrome were more prevalent amongst those who reported not using probiotics.

In the normal-weight individuals, the median glycaemic control was significantly better in the group using probiotics (Table 1). However, in overweight or obese individuals, no difference in HbA_{1c} was observed between the two groups. Of the blood lipid variables, the triglyceride concentration and the TG-HDL-cholesterol ratio were lower in the Probiotics^{yes} group. In women reporting the use of probiotics, the HDL-cholesterol concentration was higher. Finally, of the continuous outcome variables, systolic blood pressure was lower amongst those using probiotics.

No differences were observed in the dietary intake of total energy, macronutrients, and fibre between those using and not using probiotics (Table 2). Moreover, the relative distribution of fatty acid intakes was similar between the groups. However, significantly higher median sucrose intake was observed amongst those using probiotics.

Table 2: Dietary intake divided by the reported use of probiotics.

	Probiotics^{no} n=636 (61%)	**Probiotics^{yes} n=403(39%)**	**P**
Energy, kJ	7705 (6426-9219)	7677 (6371-8998)	0.745
Energy, kcal	1841 (1536-2204)	1835 (1523-2150)	0.742
Carbohydrates, E%	43 (38-48)	43 (39-48)	0.682
Sucrose, g	29.1 (20.2-48.4)	34.5 (22.2-48.9)	0.027
Proteins, E%	17 (15-19)	17 (15-19)	0.883
Fats, E%	36 (31-40)	35 (31-40)	0.252
PUFA, E%	6 (5-7)	6 (5-7)	0.774
SAFA, E%	13 ± 3	12 ± 3	0.295
MUFA, E%	12 (10-14)	12 (10-14)	0.267
Alcohol, E%	0.7 (0-3.1)	1.1 (0-3.5)	0.345

Total fibre, g	20.7 (15.8-26.4)	21.3 (16.5-27.1)	0.401
Soluble fibre, g	4.8 (3.7-6.2)	5.1 (3.9-6.5)	0.198

Data are presented as median (interquartile range) or mean ± standard deviation. Between-group comparisons with Mann-Whitney U-test or independent samples t-test, respectively. E%, percentage of total energy intake.

In the logistic regression analyses, adjusting for age, sex, nephropathy status, sucrose intake, physical activity, and smoking, the use of probiotics was associated with a 35% (P=0.006) lower odds of overweight or obesity and a 40% (P=0.003) lower odds of the metabolic syndrome (Table 3). Similarly, the odds of fulfilling the blood pressure, HDL-cholesterol, triglyceride, and waist components of the metabolic syndrome were 42% (P=0.009), 32% (P=0.025), 33% (P=0.019), and 34% (P=0.013) lower, respectively, amongst those using probiotics.

Table 3: Association between probiotic use and selected dichotomous health markers.

	Model 1				Model 2			
	B	**Exp(B)**	**95% CI for Exp(B)**	**P**	**B**	**Exp(B)**	**95% CI for Exp(B)**	**P**
OW/OB	-0.30	0.74	0.57-0.96	0.022	-0.43	0.65	0.48-0.89	0.006
MS	-0.50	0.61	0.46-0.81	0.001	-0.52	0.60	0.42-0.84	0.003
MS/BP	-0.37	0.69	0.49-0.97	0.033	-0.54	0.58	0.39-0.88	0.009
MS/HDL	-0.42	0.66	0.50-0.87	0.003	-0.38	0.68	0.49-0.95	0.025
MS/TG	-0.48	0.62	0.47-0.83	0.001	-0.40	0.67	0.48-0.94	0.019
MS/waist	-0.34	0.71	0.55-0.93	0.011	-0.41	0.66	0.48-0.92	0.013

Model 1 is adjusted for age, sex, and nephropathy status. Model 2 is further adjusted for sucrose intake, physical activity and smoking. CI, confidence interval; OW/OB, overweight or obese (BMI ≥ 25 kg/m^2); MS, metabolic syndrome; MS/BP, blood pressure component of the metabolic syndrome; MS/HDL, HDL-cholesterol component of the metabolic syndrome; MS/TG, triglyceride component of the metabolic syndrome; MS/waist, waist component of the metabolic syndrome. Logistic regression.

Of the continuous outcome variables the use of probiotics was, in the fully adjusted models, associated with lower BMI (P=0.049), waist-to-hip ratio (P=0.002), total cholesterol concentration (P=0.049), triglyceride concentration (P=0.001), and TG-HDL-cholesterol ratio (P<0.001) (Table 4). Moreover, among the normal-weight individuals, use of probiotics was associated with better glycaemic control (P=0.026), and in women it was associated with smaller waist circumference (P=0.009). However, in the multivariable models, the use of probiotics was no longer associated with systolic blood pressure or HDL-cholesterol concentration.

Table 4: Association between the use of probiotics and selected continuous health markers.

	Model 1				Model 2			
	B (95% Wald CI)	**Probiotics^{no} Mean (SE)**	**Probiotics^{yes} Mean (SE)**	**P**	**B (95% Wald CI)**	**Probiotics^{no} Mean (SE)**	**Probiotics^{yes} Mean (SE)**	**P**
HbA_{1c} (NW), mmol/mol, %	-0.31 (-0.55--0.07)	67 (1.07) 8.27 (0.08)	64 (1.35) 7.96 (0.09)	0.011	-0.31 (-0.58--0.04)	66 (1.29) 8.20 (0.10)	63 (1.47) 7.90 (0.10)	0.026
HbA_{1c} (OW/OB), mmol/mol, %	0.04 (-0.16-0.23)	67 (0.80) 8.24 (0.06)	67 (1.21) 8.28 (0.08)	0.703	0.05 (-0.17-0.28)	66 (0.90) 8.19 (0.07)	67 (1.34) 8.24 (0.09)	0.66
BMI, kg/m^2	-0.47 (-0.98-0.04)	26.2 (0.16)	25.8 (0.20)	0.07	-0.60 (-1.19--0.01)	26.3 (0.19)	25.7 (0.23)	0.049
WHR	-0.01 (-0.02--0.01)	0.89 (0.01)	0.88 (0.01)	0.037	-0.02 (-0.03--0.01)	0.89 (0.01)	0.88 (0.01)	0.002
Waist (men), cm	-2.53 (-4.84--0.22)	94.9 (0.65)	92.3 (0.98)	0.032	-1.91 (-4.60-0.79)	94.8 (0.79)	92.9 (1.12)	0.165
Waist (women), cm	-1.28 (-3.28-0.73)	85.3 (0.69)	84.1 (0.76)	0.212	-2.91 (-5.11--0.72)	85.9 (0.76)	83.0 (0.81)	0.009

SBP, mmHg	-0.43 (-2.41-1.55)	137 (0.62)	137 (0.79)	0.667	-1.94 (-4.27-0.39)	138 (0.75)	136 (0.91)	0.103
DBP, mmHg	-0.67 (-1.84- 0.50)	77 (0.37)	77 (0.47)	0.263	-0.96 (-2.37-0.45)	77 (0.45)	76 (0.55)	0.182
TC, mmol/l	-0.09 (-0.20-0.02)	4.7 (0.03)	4.6 (0.04)	0.109	-0.13 (-0.25--0.01)	4.69 (0.04)	4.56 (0.05)	0.049
HDL (men), mmol/l	0.06 (-0.03-0.14)	1.48 (0.02)	1.53 (0.04)	0.212	0.01 (-0.10-0.11)	1.52 (0.03)	1.52 (0.04)	0.947
HDL (women), mmol/l	0.07 (0.01-0.15)	1.70 (0.03)	1.77 (0.03)	0.045	0.07 (-0.01-0.16)	1.71 (0.03)	1.79 (0.03)	0.085
TG, mmol/l	-0.16 (-0.26--0.06)	1.24 (0.03)	1.08 (0.04)	0.002	-0.17 (-0.26--0.07)	1.19 (0.03)	1.02 (0.04)	0.001
TG/HDL	-0.13 (-0.21--0.04)	0.87 (0.03)	0.74 (0.03)	0.004	-0.15 (-0.23--0.06)	0.82 (0.03)	0.68 (0.03)	<0.001
hs-CRP, mg/l	-0.24 (-0.49-0.01)	1.91 (0.08)	1.67 (0.10)	0.058	-0.27 (-0.56-0.02)	1.90 (0.09)	1.63 (0.11)	0.067

Model 1 is adjusted for age, sex, and nephropathy status. Model 2 is further adjusted for sucrose intake, physical activity and smoking. Final blood pressure models are additionally adjusted for the use of antihypertensive medication; blood lipid and hs-CRP models are additionally adjusted for the use of lipid lowering medication. CI, confidence interval; SE, standard error; NW, normal-weight (BMI 25 <kg/m^2); OW/OB, overweight or obese (BMI ≥ 25 kg/m^2); BMI, body mass index; WHR, waist-to-hip ratio; SBP, systolic blood pressure; DBP, diastolic blood pressure; TC, total cholesterol; HDL, high-density lipoprotein cholesterol; TG, triglyceride; TG/HDL, triglyceride-high-density lipoprotein cholesterol ratio; hs-CRP, high-sensitivity C-reactive protein. Generalized linear model.

Discussion

In the current study, we observed that the use of probiotics-containing food products or supplements was associated with a number of beneficial health markers in type 1 diabetes. In particular, the use of probiotics was associated with lower odds of overweight or obesity. Also the odds of metabolic syndrome, and its blood pressure, HDL-cholesterol, triglyceride, and waist components were significantly lower amongst those using probiotics. Moreover, in normal-weight individuals, the use of probiotics was associated with better glycaemic control.

While, to our knowledge, this is the first study to focus on the health effects of probiotics in individuals with type 1 diabetes, a number of studies have been conducted in other populations. Of interest in diabetes, probiotics may improve glycaemic control. The exact mechanisms by which probiotics could affect glycaemia, is not known. However, it has been speculated that beneficial gut bacteria could increase the secretion of glucagon-like peptide 1 which would subsequently improve carbohydrate metabolism [28]. Other proposed mechanisms are related to improved intestinal permeability, regulation of the immune system, and reduction of Toll-like receptor 4 signalling. In their meta-analysis, Li et al. reported significant reduction in fasting blood glucose levels, following probiotics-intervention, in patients with type 2 diabetes [28]. However, no effect was observed in HbA_{1c}. As a long-term measure of glycaemia, the included intervention trials (mostly ≤ 8 weeks) may have been too short to show significant changes in HbA_{1c}. In contrast, Sun and Buys reported, in their meta-analysis of individuals with type 2 diabetes or with diabetes-associated risk factors, a significant reduction in the participants' HbA_{1c} following 6-12 weeks' probiotic intervention [29]. In their study, the glycaemia-reducing effect of probiotics was most pronounced in those with diabetes, and when multiple strains were used. Interestingly, Firouzi et al. reported, in a population of individuals with type 2 diabetes, that among normal-weight participants a 12-week probiotics-intervention resulted in greater improvements in HbA_{1c} than in those overweight or obese [27]. Encouraged by these observations, we also investigated the glycaemic control separately in normal-weight and in overweight/obese. Indeed, in the current study, the use of probiotics was associated with lower HbA_{1c} levels in the normal-weight individuals, while no association was observed amongst those with higher BMIs. While the mechanism behind this phenomenon is not known, the differences frequently observed in the Bacteroidetes to Firmicutes -ratio in the gut of obese and lean subjects, could be responsible for the observed differences. Also, it has been speculated that the dosage of probiotics, commonly used, may not be sufficient to produce beneficial effects in glycaemia in obese but, instead, weight-based dosages may be required [27].

The association between gut microbiota and obesity has been shown both in animals and humans [30]. Beyond the before-mentioned differences in the Bacteroidetes to Firmicutes ratio, it has also been shown that compared to lean, obese individuals display decreased diversity of the gut microbiota; beneficial changes take place in the composition of the fecal microbiota during weight loss interventions; and a more diverse microbial community is observed amongst subjects with greater weight loss [30]. Indeed, there is some indication that the so called "obese microbiome" has an increased capacity to harvest energy from the diet, which in due course could contribute to obesity [31]. Our results fit well with previous observations as the reported use of probiotics was favourably associated with many of the body weight-related variables.

According to one meta-analysis of randomized controlled trials in type 2 diabetes, probiotics-intervention increased HDL-cholesterol concentration, but had no significant effect on LDL-cholesterol, total cholesterol or triglyceride concentration [28]. Another meta-analysis of randomized controlled trials among subjects with high, borderline high, or normal cholesterol levels, reported significant reductions in total cholesterol and LDL-cholesterol concentrations, but found no effects in HDL-cholesterol and triglyceride concentrations [20]. There are a number of proposed mechanisms by which gut bacteria are anticipated to affect circulating lipid concentrations. Probiotic bacteria may, for example, interfere with the absorption of cholesterol from the

intestine by directly assimilating it and using it as a substrate for bacterial growth [32]. Bacteria may also deconjugate bile acids, which makes them less likely to be reabsorbed into the enterohepatic circulation [33]. Increased excretion of bile acids subsequently increases the need for de novo synthesis of cholesterol-containing bile acids, which reduces the total amount of cholesterol in the circulation. Finally, the short-chain fatty acids (SCFAs) produced by the probiotics and quickly absorbed from the intestine, may inhibit hepatic cholesterol synthesis or participate in the redistribution of cholesterol from plasma to the liver [33]. In the current study, consumption of probiotics was associated with slightly but significantly lower total cholesterol and triglyceride concentration, and also a lower triglyceride–HDL-cholesterol ratio, compared to the group not using probiotics. We were not able to find any previous reports on the association between probiotics and triglyceride–HDL-cholesterol ratio. This ratio is, however, a marker of insulin resistance. An increase in the triglyceride-HDL-cholesterol ratio is associated with parameters of cardiovascular risk, and it may predict the development of coronary heart disease and cardiovascular mortality [34]. Keeping the ratio down may, thus, be relevant also in type 1 diabetes.

The effect of probiotics on blood pressure has also been investigated, and according to a meta-analysis of such randomized controlled trials, probiotics-intervention significantly reduced both systolic and diastolic blood pressure in adult subjects with or without hypertension [19]. Importantly, interventions of a duration of at least 8 weeks were required for the observed effects. Also, those with higher blood pressure at baseline, seemed to benefit the most from the intervention, and the blood pressure-reducing effects were more evident in trials using multiple species of probiotics, as opposed to single species. Probiotics may affect blood pressure via various proposed mechanisms [19]. Gut microbiota-derived SCFAs may take part in the regulation of the renin-angiotensin system, or the probiotics may increase the absorption of nutrients and phytoestrogens with vasodilatory effects. The blood-pressure reducing effect of probiotics may also result from the reduction in adiposity. In the current study, while those reporting the use of probiotics had significantly lower systolic blood pressure, the statistical significance of this difference did not persist after controlling for confounding factors. Despite this, those reporting using probiotics had lower odds of fulfilling the blood pressure component of the metabolic syndrome. In this entity, not only systolic blood pressure, but also diastolic blood pressure and the use of antihypertensive medication are, however, taken into account.

It has been suggested that the SCFAs, produced by the beneficial gut microbes, may prevent low-grade inflammation via maintaining intestinal integrity [35]. A meta-analysis of the ability of probiotics-intervention to reduce CRP levels, in type 2 diabetes, showed an overall non-significant effect [35]. A sub-group analysis in another meta-analysis suggested, however, that a high probiotic dose, longer intervention duration, and the use of multiple strains of probiotics would, indeed, have anti-inflammatory effects [36]. In the current study, the reported use of probiotics was not associated with hs-CRP levels in type 1 diabetes. In the light of the previous reports, our observations could suggest, for example, that the use of probiotics, in this population, is sporadic rather than regular.

This study has a number of limitations that should be discussed. The cross-sectional nature of the study poses a major limitation as it prevents us from making any conclusions about causality between the use of probiotics and the health markers. Self-reporting the use of probiotics is another limitation. Not all individuals may, for example, be aware that the products they use contain probiotics, and will subsequently not report their use. Also, for some participants the reported use of probiotics, at the time of data collection, may have only been an occasional event and does not reflect a more continuous habit. Such an occasional act may not be sufficient to influence the measured variables. Based on the collected data, we cannot calculate the doses of probiotics used. Also, in many cases, the actual strain or strains used was not known. Compared to many intervention trials conducted in this field, the sample size in the current study was, however, much larger. Moreover, our study group is fairly homogenous, as participants were all diagnosed with type 1 diabetes. Finally it has to be acknowledged that more health-conscious individuals may not only consume probiotics but may also have overall healthier lifestyles that may affect the outcome variables. While we adjusted the models with factors such as physical activity, smoking, and dietary intake, we cannot rule out potential residual confounding.

In conclusion, the reported use of probiotics among individuals with type 1 diabetes, was beneficially associated with a number of health markers such as glycaemia, metabolic syndrome, obesity, and lipid parameters. Our results suggest that the use of probiotics could reduce many of the traditional risk factors related to the emergence of diabetic complications. Randomized controlled trials of sufficient duration, preferably using multiple strains of probiotics, are required in order to affirm these observations.

Conflict of interests Professor Per-Henrik Groop has received research grants from Eli Lilly and Roche, is an advisory board member for AbbVie, Astra Zeneca, Boehringer-Ingelheim, Cebix, Eli Lilly, Janssen, MSD, Medscape, Novartis, and Sanofi. He has received lecture fees from Astra Zeneca, Boehringer-Ingelheim, Eli Lilly, Genzyme, MSD, Novartis, Novo Nordisk, and Sanofi. Aila Ahola has received a research grant from Diabetes Wellness Finland. All other authors declare no conflicts of interest.

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References

1. American Diabetes Association (2017) 2. Classification and Diagnosis of Diabetes. Diabetes Care 40: S11-S24.
2. Marcovecchio ML, Lucantoni M, Chiarelli F (2011) Role of chronic and acute hyperglycemia in the development of diabetes complications. Diabetes Technology and Therapeutics 13: 389-394.
3. Lind M, Svensson AM, Kosiborod M, Gudbjörnsdottir S, Pivodic A, et al. (2014) Glycemic control and excess mortality in type 1 diabetes. The New England Journal of Medicine 371: 1972-1982.
4. Kekäläinen P, Tirkkonen H, Laatikainen T (2016) How are metabolic control targets of patients with Type 1 diabetes mellitus achieved in daily

practice in the area with high diabetes prevalence? Diabetes Research and Clinical Practice 115: 9-16.

5. Lithovius R, Harjutsalo V, Forsblom C, Saraheimo M, Groop PH (2015) The consequences of failure to achieve targets of guidelines for prevention and treatment of diabetic complications in patients with type 1 diabetes. Acta Diabetologica 52: 31-38.
6. Polsky S, Ellis SL (2015) Obesity, insulin resistance, and type 1 diabetes mellitus. Current Opinion in Endocrinology, Diabetes, and Obesity 22: 277-282.
7. Merger SR, Kerner W, Stadler M, Zeyfang A, Jehle P, et al. (2016) Prevalence and comorbidities of double diabetes. Diabetes Research and Clinical Practice 119: 48-56.
8. Groop PH, Thomas MC, Moran JL, Wadén J, Thorn LM, et al. (2009) The presence and severity of chronic kidney disease predicts all-cause mortality in type 1 diabetes. Diabetes 58: 1651-1658.
9. American Diabetes Association (2017) 4. Lifestyle Management. Diabetes Care 40: S33-S43.
10. Rydén L, Grant PJ, Anker SD, Berne C, Cosentino F, et al. (2013) ESC Guidelines on diabetes, pre-diabetes, and cardiovascular diseases developed in collaboration with the EASD: the Task Force on diabetes, pre-diabetes, and cardiovascular diseases of the European Society of Cardiology (ESC) and developed in collaboration with the European Association for the Study of Diabetes (EASD). European Heart Journal 34: 3035-3087.
11. Diabetes. Current Care Guidelines (2016).
12. Eckburg PB, Bik EM, Bernstein CN, Purdom E, Dethlefsen L, et al. (2005) Diversity of the human intestinal microbial flora. Science 308: 1635-1638.
13. Ley RE, Turnbaugh PJ, Klein S, Gordon JI (2006) Microbial ecology: human gut microbes associated with obesity. Nature 444: 1022-1023.
14. Le Chatelier E, Nielsen T, Qin J, Prifti E, Hildebrand F, et al. (2013) Richness of human gut microbiome correlates with metabolic markers. Nature 500: 541-546.
15. Kumari M, Kozyrskyj AL (2017) Gut microbial metabolism defines host metabolism: an emerging perspective in obesity and allergic inflammation. Obesity Reviews 18: 18-31.
16. Woting A, Blaut M (2016) The Intestinal Microbiota in Metabolic Disease. Nutrients 8: 202.
17. Conlon MA, Bird AR (2014) The impact of diet and lifestyle on gut microbiota and human health. Nutrients 7: 17-44.
18. Joint FAO/WHO Expert Consultation (2006) Probiotics in food: Health and nutritional properties and guidelines for evaluation.
19. Khalesi S, Sun J, Buys N, Jayasinghe R (2014) Effect of probiotics on blood pressure: a systematic review and meta-analysis of randomized, controlled trials. Hypertension 64: 897-903.
20. Guo Z, Liu XM, Zhang QX, Shen Z, Tian FW, et al. (2011) Influence of consumption of probiotics on the plasma lipid profile: a meta-analysis of randomised controlled trials. Nutrition, Metabolism, and Cardiovascular Diseases 21: 844-850.
21. Ruan Y, Sun J, He J, Chen F, Chen R, et al. (2015) Effect of Probiotics on Glycemic Control: A Systematic Review and Meta-Analysis of Randomized, Controlled Trials. PLoS One 10: e0132121.
22. Akbari V, Hendijani F (2016) Effects of probiotic supplementation in patients with type 2 diabetes: systematic review and meta-analysis. Nutrition Reviews 74: 774-784.
23. Alberti KG, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, et al. (2009) Harmonizing the metabolic syndrome: a joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity. Circulation 120: 1640-1645.
24. Ahola AJ, Mikkilä V, Mäkimattila S, Forsblom C, Freese R, et al. (2012) Energy and nutrient intakes and adherence to dietary guidelines among Finnish adults with type 1 diabetes. Annals of Medicine 44: 73-81.
25. Ahola AJ, Lassenius MI, Forsblom C, Harjutsalo V, Lehto M, et al. (2017) Dietary patterns reflecting healthy food choices are associated with lower serum LPS activity. Scientific Reports 7: 6511.
26. Wadén J, Tikkanen H, Forsblom C, Fagerudd J, Pettersson Fernholm K, et al. (2005) Leisure time physical activity is associated with poor glycemic control in type 1 diabetic women: the FinnDiane study. Diabetes Care 28: 777-782.
27. Firouzi S, Majid HA, Ismail A, Kamaruddin NA, Barakatun Nisak MY (2017) Effect of multi-strain probiotics (multi-strain microbial cell preparation) on glycemic control and other diabetes-related outcomes in people with type 2 diabetes: a randomized controlled trial. European Journal of Nutrition 56: 1535-1550.
28. Li C, Li X, Han H, Cui H, Peng M, et al. (2016) Effect of probiotics on metabolic profiles in type 2 diabetes mellitus: A meta-analysis of randomized, controlled trials. Medicine 95: e4088.
29. Sun J, Buys NJ (2016) Glucose-and glycaemic factor-lowering effects of probiotics on diabetes: a meta-analysis of randomised placebo-controlled trials. The British Journal of Nutrition 115: 1167-1177.
30. Paun A, Danska JS (2016) Modulation of type 1 and type 2 diabetes risk by the intestinal microbiome. Pediatric Diabetes 17: 469-477.
31. Turnbaugh PJ, Ley RE, Mahowald MA, Magrini V, Mardis ER, et al. (2006) An obesity-associated gut microbiome with increased capacity for energy harvest. Nature 444: 1027-1031.
32. Liong MT, Shah NP (2005) Optimization of cholesterol removal by probiotics in the presence of prebiotics by using a response surface method. Applied and Environmental Microbiology 71: 1745-1753.
33. Pereira DI, Gibson GR (2002) Effects of consumption of probiotics and prebiotics on serum lipid levels in humans. Critical Reviews in Biochemistry and Molecular Biology 37: 259-281.
34. Quispe R, Martin SS, Jones SR (2016) Triglycerides to high-density lipoprotein-cholesterol ratio, glycemic control and cardiovascular risk in obese patients with type 2 diabetes. Current Opinion in Endocrinology, Diabetes and Obesity 23: 150-156.
35. Kasińska MA, Drzewoski J (2015) Effectiveness of probiotics in type 2 diabetes: a meta-analysis. Polskie Archiwum Medycyny Wewnetrznej 125: 803-813.
36. Samah S, Ramasamy K, Lim SM, Neoh CF (2016) Probiotics for the management of type 2 diabetes mellitus: A systematic review and meta-analysis. Diabetes Research and Clinical Practice 118: 172-182.